

OPTIC-VOLUMETRIC MEASUREMENTS ON SOME HUMUS FORMS

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Introduction

Micromorphological soil studies have contributed much to our knowledge of the origin of humus forms (MÜLLER, 1887; HESSELMAN, 1929) or forest-floor types (MINDERMAN, 1960), and especially to the part played by the soil fauna and micro-flora in breaking down and converting the organic matter (amongst others: HARTMANN, 1951; JONGERIUS, 1956; JONGERIUS and SCHELLING, 1960; KUBIENA, 1948, 1953, 1955). Until now these investigations have always been of a qualitative nature. Although this allowed the various processes and phenomena to be recognized, it was at the same time virtually impossible to establish with any degree of accuracy their part and significance in the whole complex of genetic factors. Recent improvements in the technique of preparing thin sections (better quality and larger dimensions), and developments in micromorphometry, have now made it possible to describe quantitatively — i.e. in terms of volumes — the different humus forms in satisfactory detail. Three of these humus forms have been characterized in this way for the first time below.

Method

Undisturbed samples, 15 by 8 by 5 cm large, were taken from the surface soils examined. They were air-dried, and impregnated with the unsaturated polyester resin Vestopal-H. After hardening, thin sections 15 by 8 cm long and wide, and 15 microns thick were prepared from the samples.

These so-called mammoth-sized thin sections (JONGERIUS and HEINTZ-BERGER, 1963) offer big advantages over conventional preparations, only a few cm large. They usually extend over several horizons, and measurements on the features observed are much more reliable.

This last is done in two ways, namely:

(1) Point counts in a fixed grid with a Zeiss integration eyepiece I (HENNIG, 1958) under a polarizing microscope equipped with a table especially constructed for the mammoth-sized thin sections. 25 points are marked out in a grid in the eyepiece (magnifying $8\times$) to form equilateral triangles. The different components distinguished are identified below each point in the grid. The grid points are scored in terms of the categories distinguished. These counts are repeated for a large number of "sites" distributed to form the corners of equal squares on the thin section. From the total point scores for the different components their volume percentages can then be directly calculated. The number of counts to be made to meet a maximum specified margin of error can be obtained from a nomogram. Smaller components can only be counted accurately at larger magnifications. For this the section should not only be evenly thin, but it should also be free from impurities (abrasive powder). For the volumetric measurements on these three humus forms it was found necessary to count 400 different grids (= 10,000 points) on each thin section magnified $80\times$.

In this way the volumes of different diameter classes of specific components, for instance pores, can also be determined. The distance between two points in the grid in the eyepiece can be obtained for every magnification by measurement, and this allows the diameter of any component under a grid point to be estimated. With some experience very reliable results can be obtained in this manner. Bigger diameter classes cannot be measured this way however, as these often partly fall outside the visual field.

(2) Because of this limitation another method was developed to measure different pore sizes (Fig. 1). Photographic negatives 9 by 12 cm large, were made on Agfa copystat paper with a polarizing microscope equipped with a Zeiss projection attachment and cassette. In this case also the photos were taken on a fixed grid. The arrangement is basically similar to that used by KUBIENA, BECKMANN and GEYGER (1961, 1962) for the optical determination of total porosity at a particular magnification. The method of exposure is however completely different because of the different purpose here (the measurement of different pore sizes). After underexposing the photographic paper somewhat with ordinary light, the nicols are crossed and locked by means of a clamp. The paper is now exposed about 15 times as long as for the first picture, while at the same time the clamp is slowly turned. On the final picture sand grains then show up black, the finer mineral fractions and the organic matter white and the pores grey (Fig. 2).

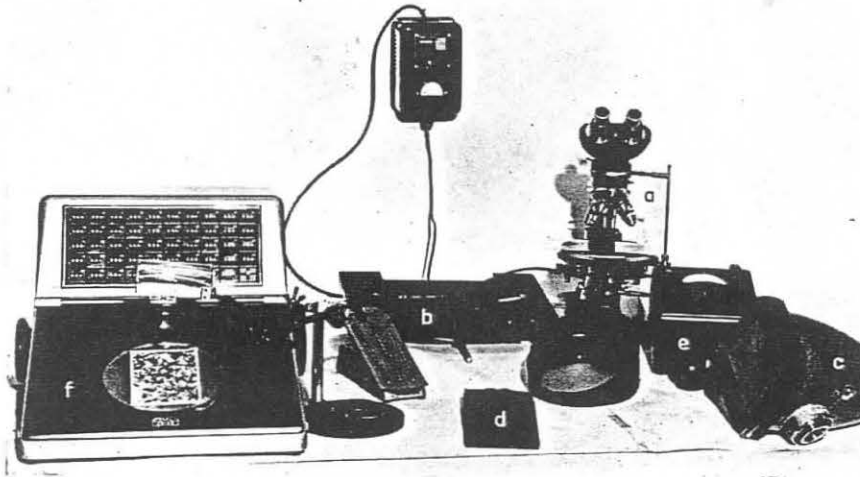


Fig. 1. The apparatus used for the measurements. (a) The polarisation microscope with 2 integration eyepieces I, and with the crossed nicols locked in position by a clamp. (b) Large mechanical stage. (c) Projection attachment with (d) cassette. (e) Photo-electric cell and exposure meter. (f) Particle size analyser.

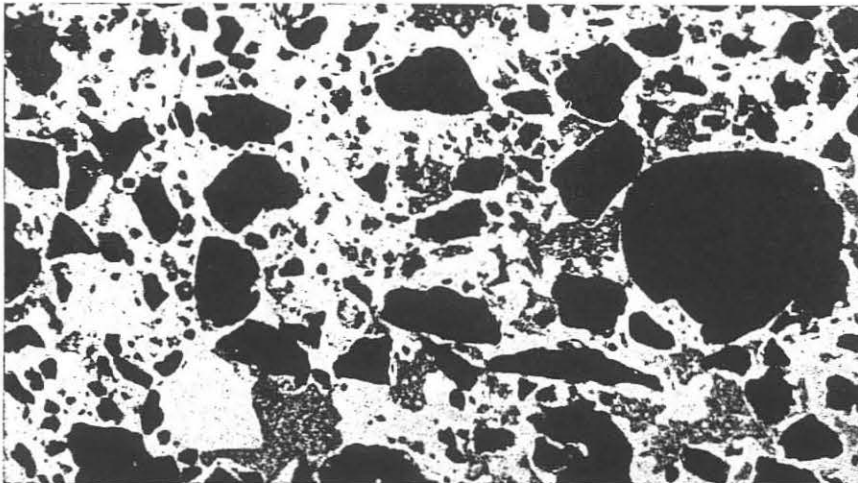


Fig. 2. An example of a photographic negative, as described in the text (True dimensions 7 by 12 cm).

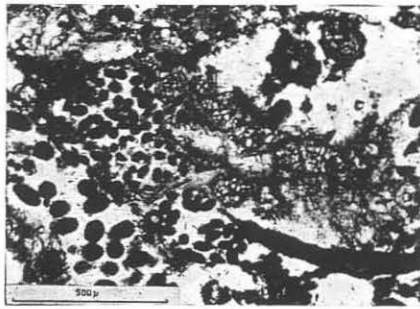


Fig. 4. Egg-shaped excrements of Oribatei amongst dark coloured leaf remains. The dark colour of the excrements is largely due to the large content of consumed fragments of fungal hyphae. Thin section of mor, A002 horizon.

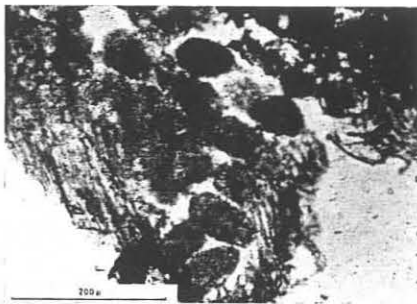


Fig. 5. Oribatei excrements, partly intact and partly merged (in the centre of the photo). Section through A002 horizon of mor.

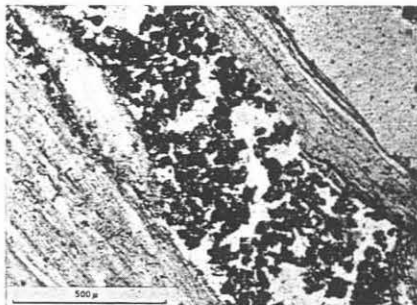


Fig. 6. Excrements of Enchytraeidae, in various stages of disintegration. Thin section of a calcareous mull, A1 horizon.

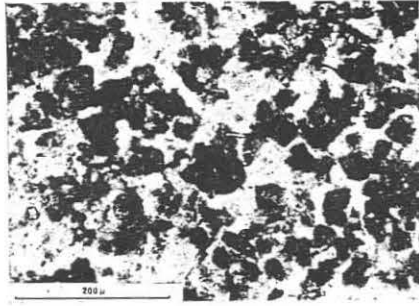


Fig. 7. Largely disintegrated excrements in the A11 horizon of a mor.

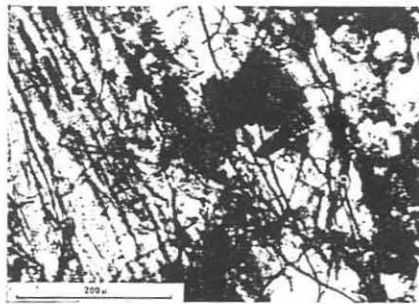


Fig. 8. Plant remains strongly penetrated by fungal hyphae. Thin section of mor, A002 horizon.

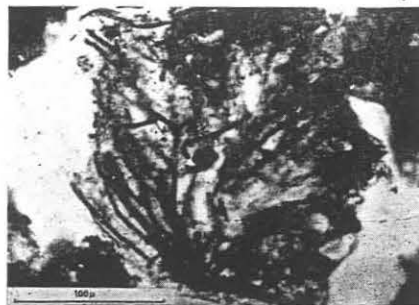


Fig. 9. Excrement strongly permeated by fungal hyphae. Thin section of mor, A002 horizon.

The photo is fixed with scotch tape to a transparent sheet marked with points to form an extended grid. This grid is constructed the same way as in the integration eyepiece I. The most common spacing between the points is half a cm, giving 446 points to a photograph of 9 by 12 cm.

Measurements of these photographs are made with a Zeiss particle size analyser. In this apparatus a light ray is passed through an iris-diaphragm to fall on a glass disc and onto the photograph placed on the latter. By adjusting the diaphragm the light-circle can be made to cover a particular

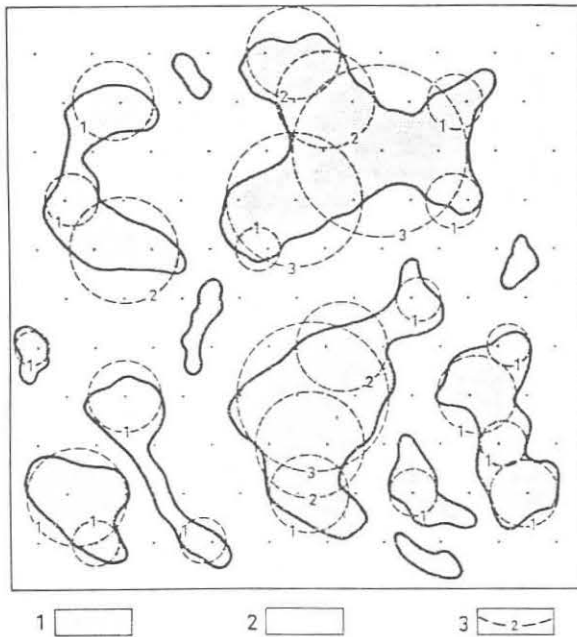


Fig. 3. The principle of measuring volumes with the particle size analyser of different pore size classes from photographs; explained in the text. 1 = pores; 2 = solid soil mass; 3 = number of grid points on the imaginary horizontal diameter of the light-circle.

particle. By pressing a foot-pedal the diaphragm diameter is recorded via a collector to a counting mechanism for 48 diameter classes (SCHLUGE, 1959). These readings however do not provide a measure of volumes. These can be obtained with the point grid. The photo is systematically moved along the (imaginary) horizontal lines connecting the gridpoints. Where a pore

covers one or more gridpoints, the diaphragm is adjusted so that the section of the line across the pore forms the diameter of the light-circle. For every gridpoint on the line section the foot-pedal is then pushed once (Fig. 3). After a photo has been completely measured this way, the different pore volumes can be calculated from the gridpoint scores.

For mammoth-sized thin sections of mineral soil, some 60 photos magnified 45 times are usually made. All pore volumes larger than about 10 microns can be measured on these.

The samples examined

These come from the ITBON experimental area at Hackfort. For a morphometric description of the humus forms, and the location of the sampling sites the reader is referred to MINDERMAN (1960).

Sample no.	Sampling site	Humus form	Horizons *)
80256	ED/18—19	Mor	A001, A002, A0, A11, A12
80259	AB/5—6	Litter-covered mull	A0, A11, A12
80252	ED/10—11	Calcareous mull with little earthworm activity	A1, A/C

*) See figs. 10 I, 11 I, 12 I. A001 = L; A002 = F; A0 = H. The distinction between the A11 and A12 horizon is based on a higher content of organic material of the A11 horizon and/or a difference in colour. In the course of preparing sample no. 80259, the A00 horizon has been lost.

The measured components

The following were measured with the integration eyepiece:

Macro- or microscopically clearly recognizable plant remains. These include litter as well as root remains. Plant remains can be fairly numerous also in the clay-humus complex. To gain an impression of the degree of microbial activity, light and dark coloured plant remains were distinguished.

Intact excrements. Three groups were distinguished:

(a) Excrements up to several mm long; cylindrical to conical (eventually weakly pointed at one or both ends), or box shaped. These are in large part produced by saprophagous macroarthropods, such as various Isopoda, Iulidae and Tipulidae, and they contain much microscopically recognizable

plant material. In the excreta from Lumbricidae, relatively fewer in number, the organic matter is more strongly comminuted.

(b) Egg shaped excrements, from 50 to 200 μm large at the most, and generally a light honey- to dark brown in colour in transmitted light; without with or only few recognizable plant remains. These come from Oribatei (Fig. 4).

(c) Excrements no larger than about 50 μm . These partly have a fairly irregular circumference, are very dark and consist of finely divided organic matter. They come for the greater part probably from Collembola and Enchytraeidae.

Disintegrating excrements. These come mainly from Oribatei and Enchytraeidae. In the first phase of disintegration they appear to expand a little (probably due to gas production by microbial activity), and lose their sharp contours. They then gradually "flow" together into small knobbly, porous aggregates, and sometimes even into an undifferentiated mass (Figs. 5, 6 and 7). The organic matter generally progressively darkens. The Collembola excrements appear very stable. Those of the macroarthropods can break down completely into their component plant parts (probably by fungal activity; Fig. 9).

Clay-humus complex. This comprises intensive mixtures of clay and very finely divided organic matter. "Clay" here also includes the loam fraction (particles $< 50\mu\text{m}$).

Fungal hyphae present in the organic matter, concentrated nearly exclusively in the cellulose-rich, recognizable plant remains (Fig. 8) and in excrements of macroarthropods (Fig. 9), and those in between (a possible measure for matting) have been counted separately.

Coarser mineral components. These are sand grains $< 50 \mu\text{m}$ and iron concretions.

The particle size analyser was used to separate the pores in the mineral horizons into 6 diameter classes: 10—30, 30—50, 50—75, 75—100, 100—200 and $> 200 \mu\text{m}$. Cell cavities in the plant remains were not counted as pores, but included with the recognizable plant remains, or with the excrements from the macroarthropods.

Results of the measurements

These are presented in Figs. 10, 11 and 12. Each figure shows:

1. A presentation of the horizons occurring in the thin section.

II. The volumes of organic matter (including the clay-humus complex), "sand" and the different porosity classes.

III. The volumes of the different components of the organic fraction per horizon.

Mor (Fig. 10). The organic matter in the A001 horizon consists nearly completely out of large, clearly recognizable litter fragments, with scattered among them a small number of intact excrements of all three categories. Because the litter in this layer is not yet matted, the total volume of the organic matter is only low (about 24%).

The A002 horizon is increasingly matted with depth. This is evident from both the increase in the volume percentage of recognizable plant remains, and from the volume of fungal hyphae. It is of interest that the amount of light coloured plant remains increases not only in absolute value, but also relative to the dark coloured ones. This results from a root concentration in this layer. The volume of excrements, both absolute and relative (with respect to the recognizable plant remains) has markedly increased. That of the excrements of the macroarthropods strongly decreases with depth, while the volume of the Oribatei excrements (which at this level contain many chewed hyphen fragments) rapidly increases. The volume of disintegrating excrements increases in a parallel manner. The amount of excreta of Collembola and Enchytraeidae remains nearly constant in this one and both the following horizons at approximately 0.4 vol. %.

In the A0 horizon the volume percentage of light- as well as dark-coloured recognizable plant remains decreases markedly. Both are present in nearly equal amounts. The volume of large excrements, found also in the top few cm of the A11 horizon increases again towards the base of the horizon. They are partly casts from Lumbricidae. It is significant that the preserved excrements from Oribatei are only small in amount while those disintegrating strongly increase. The excrements are largely dark in colour.

Because the A11 horizon is fairly densely penetrated by roots, the volume of light coloured plant parts is fairly high in most of this layer. The volume of intact Oribatei excrements is probably also high because of this. The most striking feature of this horizon is however the very large amount of disintegrating excrements (Fig. 7). In the upper part of the layer they are largely brown in colour, while with depth they become increasingly dark (progressive humification). Although it can be assumed that part of this material comes from the A00 and A0 horizons (mechanical illuviation; JONGERIUS,

1956), it seems likely that quite a significant part has been produced in the horizon itself.

In the A12 horizon the root volumes as well as the volumes of intact and disintegrating excrements clearly decrease.

Fig. 10 II shows that coarser mineral components are absent from the A00 horizon. From the A0 horizon downwards their volume gradually increases, but it remains fairly small. The peak near the boundary between the A11 and the A12 horizon is caused by a zone of iron concretions.

The pores are fairly evenly distributed throughout the whole A11 horizon, while the volume of pores $> 200 \mu\text{m}$ gradually increases with depth, because the pores in the sand skeleton have not been filled into the same extent with organic matter. The higher percentage of pores $> 200 \mu\text{m}$ near the boundary between the A11 and A12 horizons is a result of the iron zone. The fairly high volume percentage of pores between 10 and $30 \mu\text{m}$ should be noted in both mineral horizons. This favorable micro-porosity is due to the micro-aggregate structure of the disintegrating excrements.

The litter-covered mull (Fig. 11). As already mentioned the A00 horizon was unfortunately lost in the course of preparing the sample.

In the A0 horizon both the absolute and relative amounts of recognizable plant remains are fairly low and these are mostly light coloured. A relatively large proportion of the excrements comes from macroarthropods; the excrements of the other categories are present in larger amounts than in the A0 horizon of the mor. Here also the excrements of Oribatei disintegrate fairly rapidly. Neither in this horizon, nor in the two below were measurable amounts of fungal hyphae found.

The A11 horizon contains few recognizable plant remains and those are largely derived from roots. The volume of large excrements, both from macroarthropods and from Lumbricidae rapidly decreases with depth in this horizon. The second group of excrements remains fairly constant through the whole horizon, the small excrements decrease in amount. The volume of disintegrating excrements is large; humification tends to increase downwards. A homogeneous clay-humus complex — probably developed from disintegrating Lumbricidae excrements — gradually appears.

The A12 horizon is marked by a gradual decrease of the volumes of excrements and a corresponding increase of the clay-humus complex.

The volume of sand gradually increases with depth, while the pore volume decreases. It will be noted that especially the volume of pores $> 200 \mu\text{m}$

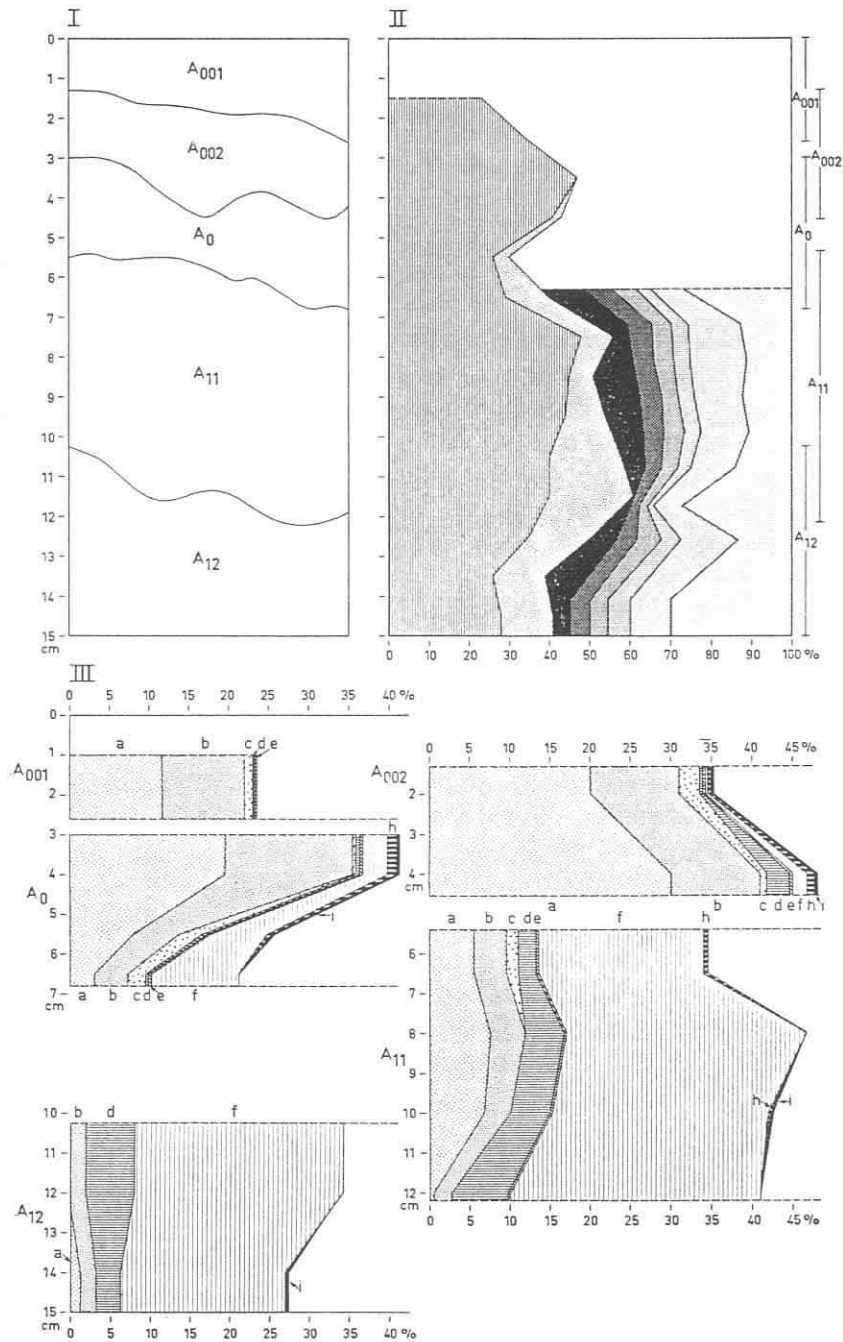


Fig. 10. The results of the volumetric measurements on mor.

decreases. Possibly this is a result of the relatively low water-stability of the clay-humus complex.

Calcareous mull with little earthworm activity (Fig. 12). The amount of recognizable plant remains, especially the dark coloured ones is even lower in the A1 horizon of this profile than in those of the previous two profiles. The volume of the excrements is also considerably lower. The volume of the very heterogeneous clay-humus complex rapidly increases with depth.

The organic matter in the A/C horizon is largely concentrated in the clay-humus complex, and the latter decreases rapidly in amount with depth: the profile proved very sandy (Fig. 12 II) and it is loamy only in the surface soil.



Here also the porosity decreases very gradually, and more particularly the volume of pores $> 200 \mu\text{m}$. The latter makes up a considerable proportion of the pores in the upper part of the A1 horizon, as many large tunnels are present here. The very small volume of pores between 10 and $30 \mu\text{m}$ should be noted as a result of the low amount of small excrements.

Conclusion







It is evident from the foregoing that the humus forms considered here show very characteristic differences in the proportions and vertical distri-

LEGEND










II

-  organic matter including clay-humus complex
-  sand $> 50\mu$ and iron concretions

Pore diameters

-  10— 30μ
-  30— 50μ
-  50— 75μ
-  75— 100μ
-  100— 200μ
-  $> 200\mu$

III

- a  light-coloured plant remains
- b  dark-coloured plant remains
- c  excrements of saprophagous macroarthropods and Lumbricidae
- d  excrements of Oribatei
- e  excrements of Collembola and Enchytraeidae
- f  disintegrating excrements
- g  clay-humus complex
- h  hyphae in plant remains and excrements
- i  hyphae between plant remains and excrements

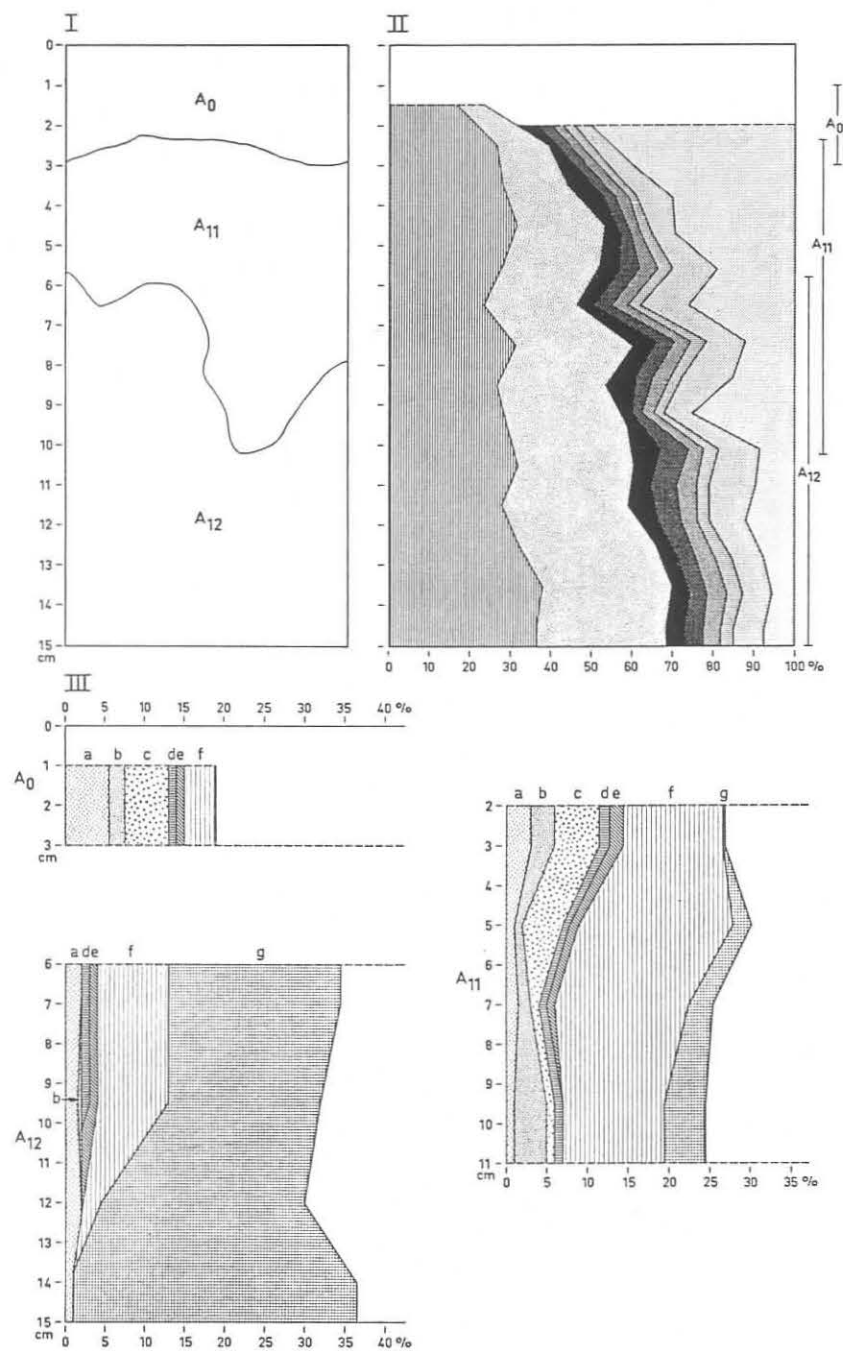


Fig. 11. The results of the volumetric measurements on a litter covered mull.

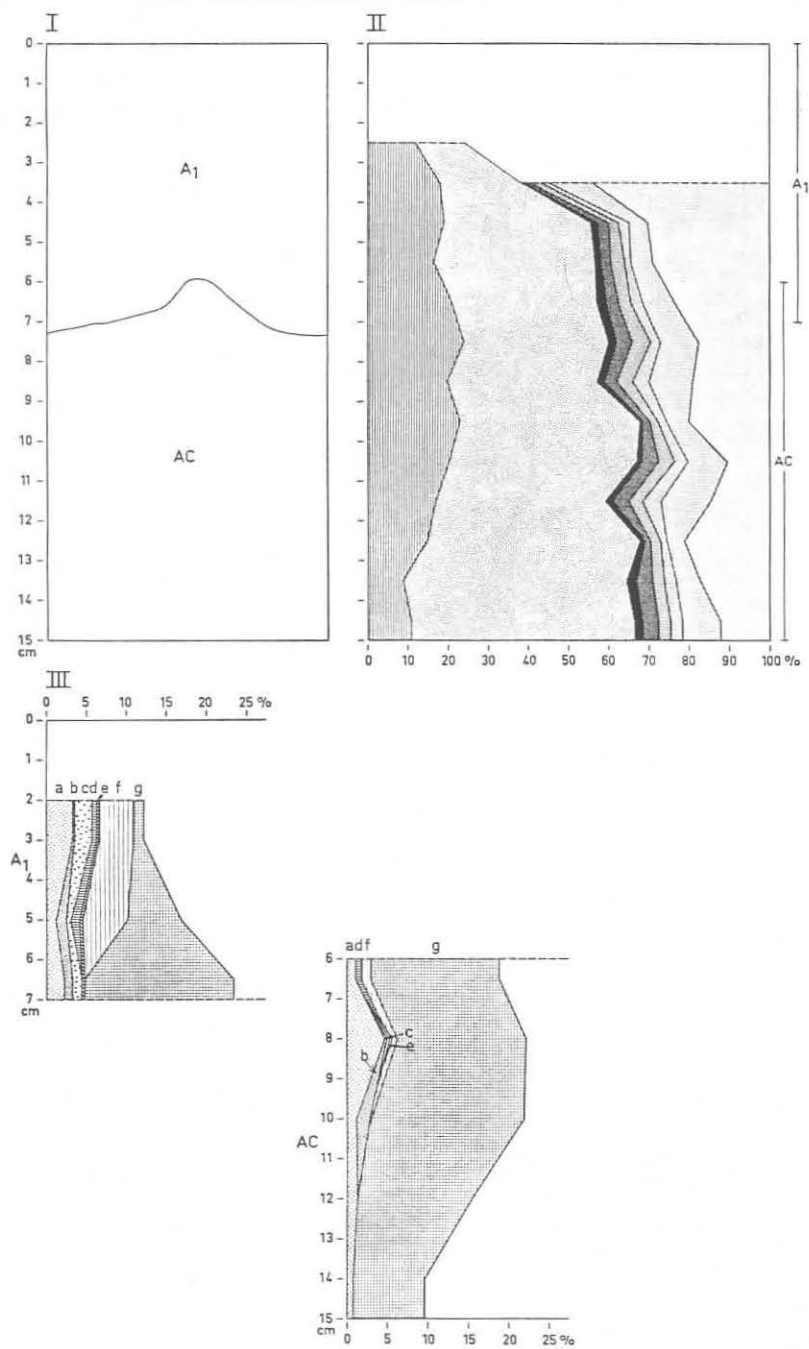


Fig. 12. The results of the volumetric measurements on a calcareous mull with little earthworm activity.

bution of the micromorphologically distinguished components. The development of a more refined classification of the excrement forms and their stages of disintegration will allow very detailed micromorphometrical analyses of the humus forms, which should extend considerably our understanding of their genesis.

Summary

Optic-volumetric measurements were carried out on "mammoth-sized" thin sections of three different humus forms (a mor, a litter-covered mull and a calcareous mull). The volumes of the different components of the organic and mineral matter were directly measured with a Zeiss integration eyepiece I. The volumes of the pore classes distinguished were determined on micro-photographs with a Zeiss particle size analyser and with the aid of a grid of fixed points. These measurements revealed very characteristic differences between the respective humus forms.

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